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(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

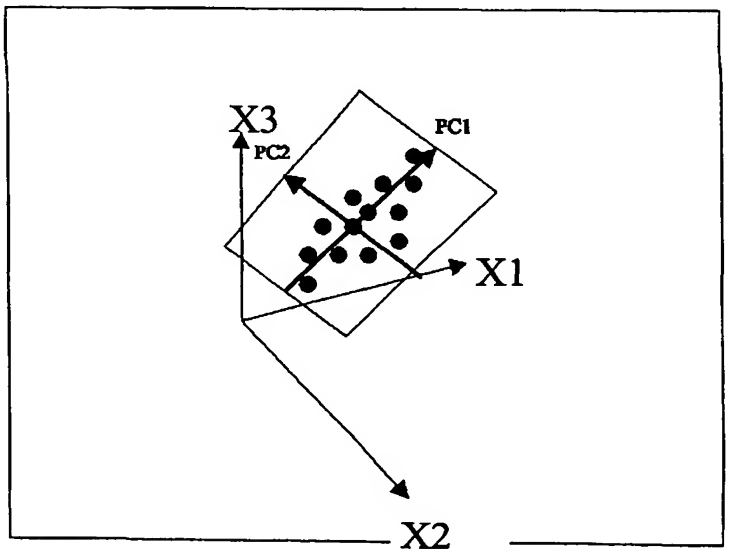
(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES,*

[Continued on next page]

(54) Title: METHOD OF RAPID DETECTION OF MUTATIONS AND NUCLEOTIDE POLYMORPHISMS USING CHEMOMETRICS



(57) Abstract: The present invention relates to methods, kits and systems for determining the presence or absence of target polynucleotides through hybridisation with polynucleotide probes comprising a detectable label and subsequent spectral analysis, preferably using multivariate analysis. The analysis of the spectral data allows to determine whether or not the probe is part of a hybrid polynucleotide and thus whether or not the target polynucleotide is present. Furthermore, the analysis of the spectral data allows to determine, when the probe is part of a hybrid polynucleotide, whether or not there is one or more mismatch between the probe and the target. The methods, kits and systems may be used for the determination of mutations and polymorphisms.

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WO 2004/024949 A3

FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG)

— *as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for all designations*

— *of inventorship (Rule 4.17(iv)) for US only*

Published:

— *with international search report*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

(88) Date of publication of the international search report:
27 May 2004

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

INTERNATIONAL SEARCH REPORT

International Application No
PCT/DK 03/00594

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12Q1/68 G06F17/10

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12Q G06F

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, MEDLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5 635 352 A (FULTZ TIMOTHY ET AL) 3 June 1997 (1997-06-03) claims 1-21 ---	4, 110-130
Y	US 6 048 690 A (O'CONNELL JAMES P ET AL) 11 April 2000 (2000-04-11) claims 1-10 ---	4, 110-130
Y	US 5 633 134 A (SHUBER TONY) 27 May 1997 (1997-05-27) claims 1-20 ---	4, 110-130
P,Y	US 6 584 413 B1 (KOTULA PAUL G ET AL) 24 June 2003 (2003-06-24) figures 6-10 --- -/--	4, 110-130

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
3 February 2004	15 MAR 2004

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Fernando Farieta

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

International Application No
PCT/DK 03/00594

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 6 261 782 B1 (FENG LI ET AL) 17 July 2001 (2001-07-17) claim 80	4, 110-130
Y	US 6 341 257 B1 (HAALAND DAVID M) 22 January 2002 (2002-01-22) column 13, line 50 - line 60	4, 110-130
A	LEE K R ET AL: "Chemometrics approach to the determination of polymorphism of a drug compound by infrared spectroscopy." DRUG DEVELOPMENT AND INDUSTRIAL PHARMACY. UNITED STATES FEB 2000, vol. 26, no. 2, February 2000 (2000-02), pages 135-147, XP002267858 ISSN: 0363-9045 figures 1-17	1-130
A	KURT VARMUZA: "Chemometrics: Statistics and Multivariate Data Analysis in Chemistry - Application Examples" INT. CONF. ON STATISTICS AND RELATED FIELDS, 5 - 9 June 2003, pages 1-29, XP002267859 Honolulu, Hawaii 2003 page 5	1-130
A	US 5 849 486 A (O'CONNELL JAMES PATRICK ET AL) 15 December 1998 (1998-12-15) claims 1-37	1-130
E	US 6 675 106 B1 (KOTULA PAUL G ET AL) 6 January 2004 (2004-01-06) claims 85-106	1-130

INTERNATIONAL SEARCH REPORT

International application No.
PCT/DK 03/00594

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 109
because they relate to subject matter not required to be searched by this Authority, namely:

see FURTHER INFORMATION sheet PCT/ISA/210

2. [X] Claims Nos.: 1-3, 25, 27, 29, 31, 33 and 35
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

see FURTHER INFORMATION sheet PCT/ISA/210

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this International application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.1

Claims Nos.: 109

Claim 109 relates to methods of treatment of the human or animal body by surgery or by therapy or diagnostic methods practiced on the human or animal body (PCT Rule 39.1(iv)). Nevertheless, a search has been executed for this claim. The search has been based on the alleged effects of the method.

Continuation of Box I.2

Claims Nos.: 1-3, 25, 27, 29, 31, 33 and 35

Present claims 1-3 relate to an extremely large number of possible methods. Support within the meaning of Article 6 PCT and / or disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the methods claimed. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible.

Claims 25, 27, 29, 31, 33 and 35
The wording "similar probes" in claims 25, 27, 29, 31, 33 and 35 is undefined. It must be evident from the claims that said similar probes are functionally equivalent, i.e. that they retain the specific activity for the related claimed methods.

Consequently, the search has been carried out for those parts of the claims which appear to be supported and disclosed, namely those parts related to the methods wherein spectral data are analyzed using multivariate analysis according to the examples 1-9 and figures 1-20.

Continuation of Box I.3

Claims Nos.: 5-108

The use of the expression "to any preceding claims" in claims 5-108 do not fulfill the requirements of PCT Rule 6.4 (a-c). However, the claims has been search to the extend it was possible.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/DK 03/00594

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5635352 A	03-06-1997	US 5681697 A	28-10-1997
		AT 241017 T	15-06-2003
		AU 694468 B2	23-07-1998
		AU 1303895 A	27-06-1995
		CA 2178598 A1	15-06-1995
		DE 69432709 D1	26-06-2003
		DE 69432709 T2	20-11-2003
		EP 0731848 A1	18-09-1996
		HU 74225 A2	28-11-1996
		JP 9507024 T	15-07-1997
		WO 9516055 A1	15-06-1995
US 6048690 A	11-04-2000	US 5849486 A	15-12-1998
		US 5632957 A	27-05-1997
		US 6017696 A	25-01-2000
		US 5605662 A	25-02-1997
		US 5849489 A	15-12-1998
		US 5565322 A	15-10-1996
		US 5532129 A	02-07-1996
		US 5787032 A	28-07-1998
		AU 7474098 A	08-12-1998
		WO 9851819 A1	19-11-1998
		US 6569382 B1	27-05-2003
		AU 723134 B2	17-08-2000
		AU 6968996 A	17-04-1997
		BR 9610618 A	06-04-1999
		CA 2233238 A1	03-04-1997
		CN 1202929 A	23-12-1998
		EP 0852617 A1	15-07-1998
		JP 11512605 T	02-11-1999
		NZ 318253 A	28-02-2000
		US 6099803 A	08-08-2000
		US 2002085954 A1	04-07-2002
		WO 9712030 A1	03-04-1997
		US 6245508 B1	12-06-2001
		US 2002155586 A1	24-10-2002
		US 2003190632 A1	09-10-2003
		US 6309602 B1	30-10-2001
		US 6375899 B1	23-04-2002
		US 6319472 B1	20-11-2001
		US 6638482 B1	28-10-2003
		US 6068818 A	30-05-2000
		US 6331274 B1	18-12-2001
		US 6225059 B1	01-05-2001
		US 6254827 B1	03-07-2001
		US 6315953 B1	13-11-2001
		US 2003073122 A1	17-04-2003
		US 2001014449 A1	16-08-2001
		US 6187642 B1	13-02-2001
		US 6306348 B1	23-10-2001
		US 6518022 B1	11-02-2003
		US 6403367 B1	11-06-2002
		US 6423271 B1	23-07-2002
		US 6540961 B1	01-04-2003
		US 6287517 B1	11-09-2001
		US 6652808 B1	25-11-2003
		US 2001026778 A1	04-10-2001
		US 2001026935 A1	04-10-2001

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/DK 03/00594

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 6048690 A		US 2001052976 A1	20-12-2001
		US 2002028503 A1	07-03-2002
		US 6051380 A	18-04-2000
		AT 220459 T	15-07-2002
US 5633134 A	27-05-1997	NONE	
US 6584413 B1	24-06-2003	NONE	
US 6261782 B1	17-07-2001	AU 6638000 A	13-03-2001
		AU 6770800 A	13-03-2001
		CA 2383264 A1	22-02-2001
		EP 1206577 A2	22-05-2002
		JP 2003527087 T	16-09-2003
		WO 0112855 A2	22-02-2001
		WO 0112856 A2	22-02-2001
		US 6403319 B1	11-06-2002
		US 6383754 B1	07-05-2002
		US 2002106649 A1	08-08-2002
		US 2003082556 A1	01-05-2003
		AU 4211500 A	23-10-2000
		CA 2367400 A1	12-10-2000
		EP 1190092 A2	27-03-2002
		JP 2002540802 T	03-12-2002
		WO 0060124 A2	12-10-2000
US 6341257 B1	22-01-2002	US 2002059047 A1	16-05-2002
		US 6415233 B1	02-07-2002
		AU 3513200 A	21-09-2000
		CA 2363417 A1	08-09-2000
		EP 1166060 A1	02-01-2002
		WO 0052433 A1	08-09-2000
US 5849486 A	15-12-1998	US 5632957 A	27-05-1997
		US 6017696 A	25-01-2000
		US 5605662 A	25-02-1997
		AU 723134 B2	17-08-2000
		AU 6968996 A	17-04-1997
		BR 9610618 A	06-04-1999
		CA 2233238 A1	03-04-1997
		CN 1202929 A	23-12-1998
		EP 0852617 A1	15-07-1998
		JP 11512605 T	02-11-1999
		NZ 318253 A	28-02-2000
		US 6099803 A	08-08-2000
		US 2002085954 A1	04-07-2002
		WO 9712030 A1	03-04-1997
		US 6245508 B1	12-06-2001
		US 2002155586 A1	24-10-2002
		US 2003190632 A1	09-10-2003
		US 6309602 B1	30-10-2001
		US 6375899 B1	23-04-2002
		US 6319472 B1	20-11-2001
		US 6638482 B1	28-10-2003
		US 6068818 A	30-05-2000
		US 6331274 B1	18-12-2001
		US 6225059 B1	01-05-2001
		US 6254827 B1	03-07-2001

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/DK 03/00594

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5849486 A		US 6315953 B1	13-11-2001
		US 2003073122 A1	17-04-2003
		US 2001014449 A1	16-08-2001
		US 6187642 B1	13-02-2001
		US 6306348 B1	23-10-2001
		US 6569382 B1	27-05-2003
		US 6518022 B1	11-02-2003
		US 6403367 B1	11-06-2002
		US 6423271 B1	23-07-2002
		US 6540961 B1	01-04-2003
		US 6287517 B1	11-09-2001
		US 6652808 B1	25-11-2003
		US 2001026778 A1	04-10-2001
		US 2001026935 A1	04-10-2001
		US 6048690 A	11-04-2000
		US 2001052976 A1	20-12-2001
		US 2002028503 A1	07-03-2002
		US 6051380 A	18-04-2000
		AT 220459 T	15-07-2002
		AU 702773 B2	04-03-1999
		AU 3507095 A	27-03-1996
		BR 9508908 A	28-10-1997
		CA 2199515 A1	14-03-1996
		CN 1164894 A ,B	12-11-1997
		DE 69527375 D1	14-08-2002
US 6675106 B1	06-01-2004	NONE	

Rec'd PCT/PTO 14 MAR 2005

PATENT COOPERATION TREATY

REC'D 31 MAR 2005
WIPO PCT

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference P548PC00	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/DK 03/00594	International filing date *(day/month/year)* 12.09.2003	Priority date *(day/month/year)* 13.09.2002
International Patent Classification (IPC) or both national classification and IPC C12Q1/68		
Applicant HVIDOVRE HOSPITAL, et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 6 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 17 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☐ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
07.04.2004	30.03.2005
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Costa Roldán, N Telephone No. +49 89 2399-7180

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/DK 03/00594

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-59 as originally filed

Claims, Numbers

1-128 received on 31.08.2004 with letter of 31.08.2004

Drawings, Sheets

1/20-20/20 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☒ contained in the international application in written form.

   ☒ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☒ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☒ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☐ the claims, Nos.:

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-107,112-115,118-128
	No:	Claims	108-111, 116,117
Inventive step (IS)	Yes:	Claims	1-107
	No:	Claims	108-128
Industrial applicability (IA)	Yes:	Claims	1-128
	No:	Claims	

2. Citations and explanations

   see separate sheet

Re Item V

Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

Reference is made to the following documents:

D1: Fellenberg K et al, PNAS, 11-09-2001, vol. 98(19), p. 10781-10786.
D2: Lee et al, Drug Development & Industrial Pharmacy, 2000, vol. 26(2), p. 135-147.
D3: Maurer M et al, Horm. Metab. Res. 2001, Vol. 33, p. 691-395.

V.1. NOVELTY (Article 33(1) (2) PCT)

V.1.1. Document **D1** (see Abstract; p. 10781 right-hand-column; fig. 2; from bridging paragraph p. 10785 to p. 10786) pertains to a method for establishing the presence of targets nucleic acids in a sample comprising multivariate analysis applied to microarray data and thus determining the presence of a target. D1 does not mention the analysis of spectral data obtained from hybridised polynucleotides and thus independent **claim 1** and dependent **claims 2 to 107** are considered novel in the meaning of Article 33 (1) PCT.

V.1.2. Independent **claim 108** does not meet the requirements of Article 6 PCT in that the matter for which protection is sought is not clearly defined. The term "instructions" is not considered a characterizing feature of a kit as it relates to its optional use.

Therefore, the kit claim 108 is only characterized by the feature of comprising at least one labelled oligonucleotide probe capable of hybridizing to a preselected region of a target polynucleotide. Such kits are absolutely standard in the art and cannot be considered novel. Therefore, **claim 108** and dependent **claims 109 to 111** also relating to instructions for the kit are **not novel.**

Kit dependent claims 112-115 are regarded as being **novel** in the meaning of Article 33 (1) PCT.

V.1.3. Claim 116 relates to a system (apparatus) used for establishing whether at least one target polynucleotide is present in a sample. Claim 116 comprises a polynucleotide probe and a target nucleotide. Said probes and targets are not considered to be an integral

part of the apparatus, and instructions are not considered a characterizing feature of an apparatus (as they relate to an optional use). Therefore, said claim pertains to an apparatus comprising a sample chamber wherein electromagnetic radiation can be recorded, means for sensing and recording electromagnetic radiations from said chamber, and multivariate data analysis. Therefore, in view of **D2** (see Abstract) **or D3** (see p. 692) . independent **claims 116 and 117** are **not novel**.

Dependent claims 118-128 are regarded as being **novel** in the meaning of Article 33 (1) PCT.

V.1.4. Therefore, **claims 1 to 107, 112 to 115 and 118 to 128** are considered **novel.**

V.2. INVENTIVE STEP (Article 33(3)PCT)

V.2.1. D1 is considered the closest prior art for claim 1.

Claim 1 differs from D1 in that spectral data is recorded and analysed.

The problem to be solved is regarded as the provision of an alternative method for detecting a target polynucleotide in a sample.

The solution proposed is the use of spectral data recorded from polynucleotide hybridisation in multivariate analysis.

Document D1 refers to multivariate analysis (i.e PCA) for analysing polynucleotide microarray data for detecting target nucleotides, however it only analyses data obtained by hybridization of radioactive labelling or fluorescent labelled polynucleotide probes.

Neither D1 nor any other prior art documents provide any indication of using spectral data obtained by polynucleotide hybridization in multivariate analysis and thus a method as disclosed in **independent claim 1 is** considered **inventive** (Article 33(3) PCT). **Dependent claims 2 to 107** are thus also considered **inventive.**

V.2.2. Dependent kit **claims 112 to 115** (additionally comprising a control, a tube container comprising linked probes) **and dependent claims 118 to 128** (pertaining to a system

comprising a computer and solid supports) seem to be routine variations in the art and do not appear to contain any additional features which, in combination of the features of any claim to which they refer, meet the requirements of the PCT with respect to inventive step (Art.33 (3) PCT).

Claims

1. A method for establishing whether at least one target polynucleotide is present in a sample, comprising the steps of

   i) adding, to a sample to be analysed for the presence of the at least one target polynucleotide, at least one polynucleotide probe at least partly complementary to a sub-sequence of the at least one target polynucleotide, wherein the at least one probe comprises at least one detectable label,

   ii) incubating the sample under conditions suitable for the formation of at least one hybrid polynucleotide comprising the at least one probe and the at least one target polynucleotide, when present,

   iii) recording spectral data from an environment comprising at least part of the sample,

   iv) analysing the spectral data using multivariate analysis, and

   v) establishing whether the target polynucleotide is present.

2. The method according to claim 1, wherein the analysis of the spectral data can distinguish for each of the at least one probe whether the probe is part of the at least one hybrid polynucleotide or not part of the at least one hybrid polynucleotide.

3. The method according to claim 1 or 2, wherein the analysis of the spectral data can distinguish for each of the at least one probe, when the probe is part of the at least one hybrid polynucleotide, whether or not there is a mismatch between the probe and the sub-sequence of the at least one target polynucleotide.

4. The method according to any of the preceding claims 1 to 3, wherein the at least one probe has a length of 6 to 50 nucleotides, preferably 6 to 25 nucleotides, such as 6 to 8 nucleotides, 8-10 nucleotides, 10-12 nucleotides, 12-14

nucleotides, 14-16 nucleotides, 16-18 nucleotides, 18-20 nucleotides, 20-22 nucleotides, or 22-25 nucleotides.

5. The method according to claim 4, wherein the sequence complementarity between target and probe in a range of overlap is at least 50%, more preferably at least 60 %, more preferably at least 70 %, more preferably at least 80 %, more preferably at least 85 %, more preferably at least 90 %, more preferably at least 95 %, more preferably at least 96 %, more preferably at least 98 %, more preferably 100%.

6. The method according to any of the preceding claims, wherein at least one probe comprises at least one RNA monomer.

7. The method according to any of the preceding claims, wherein at least one probe comprises at least one DNA monomer.

8. The method according to any of the preceding claims, wherein at least one probe comprises at least one PNA monomer.

9. The method according to any of the preceding claims, wherein at least one probe comprises at least one methylated monomer.

10. The method according to any of the preceding claims, wherein at least one probe comprises at least one LNA monomer.

11. The method according to any of the preceding claims, wherein at least one probe comprises a mixture of monomers in claims 6-10.

12. The method according to any of the preceding claims, wherein one probe is capable of hybridising to two or more target polynucleotides.

13. The method according to any of the preceding claims, comprising using at least two polynucleotide probes capable of hybridising to two different target polynucleotides.

14. The method according to any of claims 1 to 11, comprising using at least two polynucleotide probes capable of hybridising to the same target polynucleotide.

15. The method according to claim 13 or 14, wherein the two probes are linked to identical or different detectable labels.

16. The method according to any of the claims 1 to 11, comprising using at least three probes capable of hybridising to three different target polynucleotides.

17. The method according to any of claims 1 to 11, comprising using at least three probes capable of hybridising to one or two different target polynucleotides

18. The method according to claim 16 or 17, wherein the three probes are linked to identical or different detectable labels.

19. The method according to any of claims 1 to 11, comprising using at least four oligonucleotide probes capable of hybridising to four different target polynucleotides.

20. The method according to any of claims 1 to 11, comprising using at least four oligonucleotide probes capable of hybridising to one, two or three different target polynucleotides.

21. The method according to claim 19 or 20, wherein the four labels are linked to identical or different detectable labels.

22. The method according to any of claims 1 to 11, comprising using five or more probes capable of hybridising to different target polynucleotides.

23. The method according to any of the preceding claims, wherein at least one probe comprises a probe selective for apolipoprotein B mutations related to atherosclerosis.

24. The method according to claim 23, wherein the probe comprises a sequence from any of SEQ ID NO 1 to 4 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

25. The method according to any of the preceding claims, wherein at least one probe is selective for apolipoprotein E polymorphism (apoE2, E3 and E4) related to neurological diseases.

26. The method according to claim 25, wherein the probe comprises a sequence from any of SEQ ID NO 5 to 8 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

27. The method according to any of the preceding claims, wherein at least one probe is selective for human muscle glycogen synthase polymorphism related to diabetes mellitus.

28. The method according to claim 27, wherein the probe comprises a sequence from any of SEQ ID NO 9 to 10 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

29. The method according to any of the preceding claims, wherein at least one probe is selective for methylene tetrahydrofolate reductase polymorphism related to osteoporose.

30. The method according to claim 29, wherein the probe comprises a sequence from any of SEQ ID NO 13 to 14 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

31. The method according to any of the preceding claims, wherein at least one probe is selective for Dnase1 mutations related to rheumatological diseases.

32. The method according to claim 31, wherein the probe comprises a sequence from any of SEQ ID NO 11 to 12 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

33. The method according to any of the preceding claims, wherein at least one probe is selective for a mutation in the BRCA1 gene or in the BRCA2 gene.

34. The method according to claim 33, where the probe comprises a nucleotide sequence selected from any of SEQ ID No 27-30 or similar probes, where the mutation/polymorphism is placed in the 5' or 3' end.

35. The method according to any of the preceding claims, wherein at least one probe is selective for mismatch repair gene mutations related to cancer.

36. The method according to claim 35, wherein the probe comprises a sequence from any of SEQ ID NO 15 to 16.

37. The method according to any of the preceding claims, wherein at least one probe is selective for a mutation in a promoter sequence or other expression signal.

38. The method according to any of the preceding claims, wherein at least one probe is selective for a mutation in a coding sequence (exons) and the intervening introns.

39. The method according to any of the preceding claims, wherein at least one probe is selective for a microbial target nucleic acid sequence.

40. The method according to claim 39, wherein the probe is selective for a microbial 16S, 18S, or 23S rRNA sequence.

41. The method according to claim 40, wherein the probe has a nucleotide sequence selected from SEQ ID NO 17 to 19.

42. The method according to any of the preceding claims, wherein at least one target polynucleotide comprises RNA, such as mRNA and/or rRNA and or tRNA

43. The method according to claim 42, wherein the rRNA comprises 5S, 5.5-5.8S, 16S, 18S, 23S, 25-28S rRNA.

44. The method according to any of the preceding claims, wherein at least one target polynucleotide comprises DNA.

45. The method according to claim 44, wherein the DNA is selected from the group comprising genomic DNA, organelle DNA, mitochondrial DNA, chloroplast DNA, cDNA, environmental DNA, virus DNA.

46. The method according to any of the preceding claims, wherein at least one target polynucleotide comprises a synthetic polynucleotide sequence.

47. The method according to any of the preceding claims, further comprising inclusion of various control polynucleotides in the hybridisation mixture, such as positive controls (wild-type, mutation, heterozygote), negative control (dummy DNA sequence).

48. The method according to any of the preceding claims, wherein the target polynucleotide comprises chemically or biologically modified nucleic acids.

49. The method according to claim 48, wherein the modification comprises modification of cytosine by bisulphite.

50. The method according to any of claims 42 to 49, wherein the target polynucleotide comprises a mixed polymer of any of the polymers of claims 42 to 49.

51. The method according to any of the preceding claims, wherein at least one target polynucleotide has a length of 8 bases to 1000 kb.

52. The method according to claim 51, wherein the length of at least one target polynucleotide is from 8-15 bases, from 15-30 bases, from 30 to 50 bases, from 50 to 100 bases, from 100 to 200 bases, from 200 to 300 bases, from 300 to 500 bases, from 500 to 750 bases, from 750 to 1000 bases, from 1000 to 1500 bases, from 1500 to 3000 bases, from 3000 to 5000 bases, from 5000 to 10000 bases, from 10000 to 15000 bases, from 15000 to 20000 bases, from 20000 to

25000, from 25000 to 30000 bases, from 30000 to 35000 bases, from 35000 to 40000 bases, from 40000 to 45000 bases, from 45000 to 50000 bases, from 50000 to 75000 bases, from 75000 to 100000 bases, from 100 kb to 250 kb, from 250 to 500 kb, from 500 kb to 750 kb, from 750 kb to 1000 kb, or more than 1000 kb.

53. The method according to any of the preceding claims, wherein the length of the overlap between the probe and target polynucleotide is at least 5 nucleotides, more preferably at least 6 nucleotides, such as at least 7 nucleotides, for example 8 nucleotides, such as at least 9 nucleotides, for example at least 10 nucleotides, such as at least 15 nucleotides, for example at least 20 nucleotides, for example at least 25 nucleotides, such as at least 50 nucleotides, for example at least 100 nucleotides.

54. The method according to any of the preceding claims, wherein the length of at least one probe is 7 to 1000 nucleotides, such as from 7 to 10 nucleotides, 10 to 15 nucleotides, 15 to 20 nucleotides, 20 to 25 nucleotides, 25 to 35 nucleotides, 35 to 50 nucleotides, 50 to 75 nucleotides, 75 to 100 nucleotides, 100 to 150 nucleotides, 150 to 200 nucleotides, 200 to 250 nucleotides, 250 to 350 nucleotides, 350 to 500 nucleotides, 500 to 750 nucleotides, 750 to 1000 nucleotides, or above 1000 nucleotides.

55. The method according to any of the preceding claims, wherein the nucleotide being complementary to a polymorphism/mutation in a target polynucleotide is positioned in the 3' or 5' terminal of the probe.

56. The method according to any of the preceding claims 1 to 54, wherein the nucleotide being complementary to a polymorphism/mutation in a target polynucleotide is positioned in the centre of the probe.

57. The method according to any of the preceding claims 1 to 54, wherein the nucleotide being complementary to a polymorphism/mutation in the target polynucleotide is positioned at least 1 nucleotide from the 3' or 5' terminal, such as at least 2 nucleotides from the 3' or 5' terminal, for example at lest 3

nucleotides from any of said terminals, such as at least 5 nucleotides from any of said terminals, for example at least 10 nucleotides from any of said terminals.

58. The method according to any of the preceding claims 1 to 54, wherein the probe comprises a sequence which is complementary to the sequence lying immediately upstream or immediately downstream to a polymorphic site in the target polynucleotide and the probe does not contain a nucleotide being complementary to the polymorphic site.

59. The method according to any of the preceding claims, wherein at least one label is bound to the 3' or 5' terminal nucleotide of the probe.

60. The method according to any of the preceding claims, wherein at least one label is bound to a non-terminal nucleotide of the probe.

61. The method according to any of the preceding claims, wherein at least one label is bound to the nucleotide being complementary to the polymorphic site.

62. The method according to any of the preceding claims, wherein at least one label is bound to a nucleotide at least 1 nucleotide upstream or downstream to the nucleotide complementary to the polymorphic site, such as at least 2 nucleotides upstream or downstream, for example at least 3 nucleotides, such as at least 5 nucleotides, for example at least 10 nucleotides.

63. The method according to any of the preceding claims, wherein at least one probe has at least two stretches of complementarity to at least one target polynucleotide, such as at least 3 stretches, for example at least 4 stretches, such as at least 5 stretches.

64. The method according to claim 63, wherein two stretches are separated by a nucleotide sequences, which does not hybridise to the target polynucleotide.

65. The method according to any of the preceding claims, further comprising amplification of a polynucleotide prior to hybridisation.

66. The method according to claim 65, wherein the amplification comprises PCR, long range PCR, and any variant of PCR amplification.

67. The method according to claim 65, wherein the amplification comprises ligase chain reaction, asymmetric amplification, single-strand amplification, T7 amplification, NASBA (Nucleic Acid Sequence-Based Amplification), strand displacement amplification, or rolling circle amplification, or T7 polymerase amplification.

68. The method according to claim 65, wherein the amplification comprises amplification in bacteria, yeast, other cells, YAC amplification, BAC amplification or other artificial chromosome based amplifications.

69. The method according to claim 65, wherein the amplification comprises allele specific amplification.

70. The method according to any of the preceding claims, wherein undesired hybridisation reactions are prevented by the addition of one or more helper polynucleotides capable of hybridising to the target polynucleotide at a sub-sequence which does not overlap with the sub-sequence to which the probe hybridises.

71. The method according to any of the preceding claims, wherein prior to the hybridisation, a step aimed at generating single stranded polynucleotides is performed.

72. The method according to any of the preceding claims, wherein the formation of a hybrid polynucleotide takes place under conditions of

a) optimal or suboptimal stringency providing sufficient stable complexes for discriminatory signal detection,

b) any composition of buffers optimising discriminatory signal detection,

c) any form and concentrations of one or more salts optimising discriminatory signal detection,

d) any additives including but not limited to stabilisers and/or quenchers optimising discriminatory signal detection,

e) temperature range for hybridisation specific for any specific combination of analyte and probe optimising discriminatory signal detection, and/or

f) any range of time of hybridisation necessary to optimise discriminatory signal detection.

73. The method according to any of the preceding claims, wherein the formation of a hybrid is performed at a temperature between 10 and 90°C such as 10 to 20 °C, 20-30 °C, 30 to 40 °C, 40 to 50 °C, 50 to 60 °C, 60 to 70 °C, 70 to 80°C, or 80 to 90°C.

74. The method according to any of the preceding claims, wherein the formation of a hybrid is performed in a buffer, which is a PCR buffer, and/or which is non-fluorescent, and/or which stabilises the spectrum of electromagnetic radiation, and/or which allows hybridisation.

75. The method according to any of the preceding claims, wherein hybridisation is carried out under conditions of high stringency allowing hybridisation only between perfect matches.

76. The method according to any of the preceding claims, wherein hybridisation is carried out under conditions of medium to high stringency allowing hybridisation between probe and target in the presence of one or more mismatches.

77. The method according to any of the preceding claims, wherein hybridisation is carried out in solution.

78. The method according to any of the preceding claims, wherein the target or the probe is linked to a solid support prior to hybridisation.

79. The method according to claim 78, wherein said solid support comprises beads such as magnetic beads and/or the surface of a well.

80. The method according to any of the preceding claims, wherein at least one probe hybridises only to one target polynucleotide.

81. The method according to any of the preceding claims, wherein at least one probe hybridises to both a wild-type target polynucleotide and to a target polynucleotide carrying a mutation or polymorphism.

82. The method according to any of the preceding claims, wherein the at least one detectable label comprises a fluorescent label.

83. The method according to claim 82, wherein the label is selected from the list in table 2 and 3.

84. The method according to any of the preceding claims, wherein the at least one label comprises a phosphorescent label.

85. The method according to any of the preceding claims, wherein the at least one label comprises a chromogenic label such as TMB (3,3',5,5-tetramethylbenzidine).

86. The method according to any of the preceding claims, wherein recording spectral data comprises detection of signal for at least 10 discrete wavelengths, more preferably at least 20 discrete wavelengths, more preferably at least 50 discrete wavelengths, more preferably at least 100 discrete wavelengths, such as at least 200 discrete wavelengths, for example at least 250 discrete wavelengths, such as at least 300 discrete wavelengths, for example at least 400 discrete wavelengths, such as at least 500 discrete wavelengths, for example at least 600 discrete wavelengths, such as at least 750 discrete wavelengths, for example at least 1000 discrete wavelengths, such as at least 1250 discrete wavelengths, for example at least 1500 discrete wavelengths, such as at least 2000 discrete wavelengths.

87. The method according to claim 86, wherein the distance between the discrete wavelengths is 10 nm or less, more preferably 5 nm or less, more preferably 3 nm or less, more preferably 2 nm or less, more preferably 1 nm or less, such as 0.8 nm, for example 0.75 nm, such as 0.7 m, for example 0.6 nm, such as 0.5 nm, for example 0.25 nm, such as 0.1 nm, for example 0.05 nm or less, such as 0.01 nm or less.

88. The method according to any of the preceding claims, wherein the spectral data recorded comprises a fluorescence spectrum between 180 and 950 nm.

89. The method according to claim 88, wherein the fluorescence spectrum is an excitation spectrum.

90. The method according to claim 88, wherein the fluorescence spectrum is an emission spectrum.

91. The method according to any of the preceding claims, further comprising recording of spectral data from the polynucleotide probe alone.

92. The method according to any of the preceding claims, further comprising recording spectral data from the hybrid polynucleotide and from a polynucleotide probe alone and/or, from a non-hybridising polynucleotide probe contacted by the target polynucleotide, and/or from a polynucleotide probe contacted with a non-hybridising polynucleotide sequence.

93. The method according to any of the preceding claims, wherein multivariate analysis comprises general multivariate analysis, principal component analysis and extensions of this, exploratory and confirmatory factor analysis in its various forms, Cluster and latent class analysis including scaled latent class analysis, structural equation analysis, Fixed mixture analysis and combinations hereof.

94. The method according to any of the preceding claims, wherein data are treated using a neural network.

95. The method according to any of the preceding claims, wherein spectral data are recorded from hybrid polynucleotides in solution.

96. The method according to claim 95, wherein the spectral data are recorded from a solution comprising both the hybrid polynucleotide and unhybridised probe.

97. The method according to any of the preceding claims 1 to 94, wherein spectral data are recorded from hybrid polynucleotides bound to a solid support.

98. The method according to claim 97, wherein the solid support comprises a solid surface capable of immobilising a capture probe, a capture probe capable of immobilising the target polynucleotide, and a labelled detection probe capable of hybridising to the immobilised target polynucleotide.

99. The method according to claim 97, wherein the solid support is a disposable or reusable device such as but not exclusively a flow-through system.

100. The method according to claim 97, wherein the capture probe is immobilised a priori to the solid surface.

101. The method according to claim 97, wherein the capture probe is hybridised to the target before immobilisation on a solid support.

102. The method according to claim 97, wherein the capture probe(s) is/are (an) allele specific probe(s).

103. The method according to any of the preceding claims 1 to 94, wherein spectral data are recorded from hybrid polynucleotides in a gas phase.

104. The method according to any of the preceding claims 1 to 94, wherein spectral data are recorded from hybrid polynucleotides in vacuum.

105. The method according to any of the preceding claims, wherein the spectral data are recorded via mass spectroscopy.

106. The method according to any of the preceding claims, further comprising the step of determining the presence or absence of a mutation or polymorphism in the genome of an individual on the basis of the information obtained concerning the presence or absence of the at least one target polynucleotide.

107. The method according to any of the preceding claims, further comprising the step of diagnosing a disease or health related state or determining a genetic predisposition of an individual on the basis of the information obtained concerning the presence or absence of the at least one target polynucleotide.

108. A kit for detection of a mutation or a polymorphism comprising

at least one oligonucleotide probe capable of hybridising to a preselected region of a target polynucleotide, the polynucleotide probe further comprising at least one detectable label,

instructions enabling correlation of spectral data recorded from a hybrid polynucleotide between said at least one oligonucleotide probe and said target polynucleotide to the presence or absence of said mutation or polymorphism using multivariate analysis.

109. The kit according to claim 108, wherein the instructions are in the form of calibration data on a data carrier, such as floppy disc, a CD-ROM, a DVD, ROM, chips, memory-cards, bar-codes.

110. The kit according to claim 108, wherein the instructions are in the form of the address of a propagated signal comprising calibration data, which can be transferred over a network, such as e-mail, internet, on-line nets, fibre-optics, power-cables, satellite-dishes.

111. The kit according to claim 108, wherein the instructions are in the form of calibration data which can be entered into a computer unit.

112. The kit according to claim 108, further comprising at least one control polynucleotide capable of hybridising to the oligonucleotide probe and non-hybridising polynucleotide(s)

113. The kit according to claim 108, being in the form of a tube container with at least one probe linked to the inner surface, being a solid surface, the tube wall allowing electromagnetic radiation to pass the walls.

114. The kit according to claim 113, wherein the tube comprises more than one probe linked to more than one location, the locations being spatially separate.

115. The kit according to claim 113, wherein the tube comprises more than one probe the probes having detectably different labels.

116. A system for establishing whether at least one target polynucleotide is present in a sample, comprising

i) at least one polynucleotide probe being at least complementary to a target polynucleotide, the probe comprising a detectable label,

ii) a sample chamber from which electromagnetic radiation can be recorded,

iii) a source of spectrally resolved electromagnetic radiation,

iv) means for sensing and recording a spectrum of electromagnetic radiation from the sample chamber, and

v) a computer unit for storing spectral data of electromagnetic radiation and having instructions to treat the recorded spectral data using multivariate analysis.

117. A system for detection of a hybrid polynucleotide comprising

i) at least one oligonucleotide probe being at least partly complementary to a target polynucleotide, the probe comprising a detectable label,

ii) a sample chamber from which electromagnetic radiation can be recorded,

iii) a source of spectrally resolved electromagnetic radiation,

iv) means for sensing and recording a spectrum of electromagnetic radiation from the sample chamber, and

v) a computer unit for storing spectral data of electromagnetic radiation and having instructions to treat the recorded spectral data using multivariate analysis.

118. The system according to claim 116 or 117, further comprising a computer controlled robot to transfer solutions to the sample chamber.

119. The system according to claim 116 or 117, further comprising means to control the temperature of the sample chamber.

120. The system according to claim 116 or 117, wherein the sample chamber is in the form of a tube with at least one probe linked to the inner surface.

121. The system according to claim 120, wherein the tube comprises more than one probe linked to more than one spatially separate location and the system comprises means to record a spectrum from each of the spatially separate locations.

122. The system according to claim 120, wherein the tube comprises more than one probe, the more than one probe having detectably different labels.

123. The system according to claim 116 or 117, being adapted to accommodate a multi-well dish and record a spectrum for each well.

124. The system according to claim 123, being adapted to accommodate a 96 well dish.

125. The system according to claim 123, being adapted to accommodate a 384 well or more dish.

126. The system according to claim 123, being adapted to accommodate a solid support such as but not exclusively a dish or a rod.

127. The system according to claim 123, being adapted to accommodate spinning dishes or rotating and displaceable rods.

128. The system according to any of claims 116 to 127, wherein the sample chamber further comprises means for immobilisation of one or more target polynucleotides.

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